

JPRS: 18,589

OTS: 63-21528

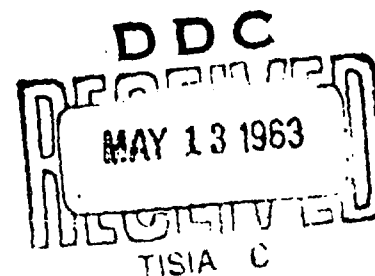
8 April 1963

AD No. 405 239

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SOVIET STUDIES OF SPACE EMBRYOLOGY

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U. S. DEPARTMENT OF COMMERCE  
OFFICE OF TECHNICAL SERVICES  
JOINT PUBLICATIONS RESEARCH SERVICE  
Building T-30  
Ohio Dr. and Independence Ave., S.W.  
Washington 25, D. C.

Price: \$.50

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SOVIET STUDIES OF SPACE EMBRYOLOGY  
(SPACE EMBRYOLOGY)

Following is the translation of an article by Doctor of Biological Sciences A. Neyfakh in the Russian-language publication Nauka i Zhizn' (Science and Life), No 1, January, 1963, Moscow, pages 65-67.]

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One of the problems of space biology is the problem of the development of an organism during space flight. The purpose of studying this problem is not single but double, two aspects as it were opposed to each other, but in reality supplementing each other. The first is the investigation of the conditions of space flight by means of a developing embryo, and the second -- investigation of the mechanism of embryonic development by means of space flight conditions.

In the flight into space man anticipates a whole complex of environmental conditions new and never encountered on earth: overloading and vibrations during liftoff and return to earth, weightlessness, and radiation during the flight period. We are able to model many of these on earth, many can be measured during the time of the flight by means of instruments, however, to know what effect all these conditions have on the organism in totality is possible to us only through biological experiments. It is not mere chance that each step of man into the cosmos was prefaced with a study of animals and plants. Neither is the very selection of these organisms which in this case serve as unique living dosimeters left to mere chance. Their appropriate selection affords a precise evaluation of the role of a given factor or of the totality of factors. Animal embryos can serve as especially sensitive dosimeters. This is due to the fact that the damage which the embryo undergoes, since it consists of the early stages of development from a few cells, is subsequently passed on to many cells of the growing organism and thus the injury is intensified many-fold. Radiation damage, for example, to the chromosomes in the egg nucleus during the course of its further division is passed on to all the cells of the embryo and leads to the formation of a malformed or nonviable organism. Thus, radiation damage in the organism of an adult mouse can be discovered only after the action of doses of 100-200 roentgens. In order to bring about the certain death of the animal not less than 700 roentgens are necessary. At the

same time, upon irradiating a mouse embryo noticeable damage is visible even at doses of 25-40 roentgens and death of the embryo occurs at doses of 100-200 roentgens. Upon irradiation of the egg the action of such small doses as 5-10 roentgens can be detected. Consequently, developing embryos can be used in space flight as a particularly sensitive biological dosimeter of radiation danger.

There is still another task whose solution compels us to know how development occurs under space flight conditions. We refer here to the creation of a closed biological cycle. It is well known that to provide cosmonauts with oxygen and food over a long flight it is necessary to set up a system in which by means of solar energy decay products would be reformed into oxygen and usable food substance. One of the links of such a cycle must necessarily be plants, which assimilate solar energy and use carbon dioxide for biological synthesis, releasing oxygen in exchange. However, plant proteins cannot always be directly assimilated by man. As a link standing between plants and man, we can certainly turn to some rapidly developing and multiplying animals.

Let us now look at the second purpose -- the use of space flight to study the mechanisms of development, to solve embryological and general-biological problems. Overloading, vibration, and radiation can be relatively easily reproduced on earth, and the study of their effect on the embryo in an artificial satellite is of no great distinction. The truth is that on earth we have not yet been able to produce such high energies as are seen in primary cosmic rays. The study of the characteristics of their action on the organism, including the embryonic organism, is of definite interest for the radiobiologists. There is still another factor of the space flights which cannot be reproduced and used in practice on earth -- this is weightlessness. It is precisely its effect on the course of embryonic development that is of special interest.

The successful flights of our cosmonauts have shown that man can survive a period of residence in the cosmos without damage, including also the condition of weightlessness. But are we certain that the embryo can easily survive weightlessness and that the earth's gravity is not a necessary condition for development? This is not at all as clear. How, for example, will the chicken egg develop under conditions of weightlessness? It would appear, normally. The embryo is enclosed in its shell, and it is more rapidly damaged by overloading than by weightlessness. However, although an experiment has thus far not been made, it can be anticipated. We refer here to the fact that in the early stages of development the chicken embryo lies very snugly against the inner wall of its shell: it is found above the yolk, and the yolk itself being lighter than the white of the egg lies against the upper inner surface of the egg. This promotes a rapid diffusion of the gases -- oxygen and carbon dioxide -- to and from the embryo. Under conditions of weightlessness the yolk can be in any position and the embryo itself

can lie on any side of the yolk. Access to oxygen will be clearly made difficult, and this can prove fatal to the embryo. (At later stages of development the respiration of bird embryos is accomplished by means of vessels lining the egg on the inside.) We presented this example only for an illustration. If the chicken embryo actually "suffocates" in the condition of weightlessness then the mechanism of this disturbance is clear and is not of great scientific interest.

More complex is the problem of the emergence of bilateral (two-sided) symmetry in development, and the role of the earth's gravity in this process. This must be discussed in more detail. And this compels us to depart from "space biology" for a while and examine a purely earthly embryological problem.

Most of the animals on earth are bilaterally symmetrical: their body can be divided by a plane into left and right halves, their dorsal and abdominal sides can be differentiated, as well as the head and caudal ends of the body. Only a few of the most simply organized and usually slow-moving animals have a radial symmetry in place of a bilateral: we can distinguish in these animals only a single axis of the body, along which the same organs are radially situated. These animals have only a dorsal (or upper) and abdominal (or lower) side, but no head or caudal ends. Typical representatives of radially symmetrical animals are, for example, the medusa or the hydra.

When and how does the plane of bilateral symmetry and its two constituent axis -- from top to bottom and from forward to the rear -- emerge in animals? The egg as a rule is radially symmetrical: it has only a single axis, usually leading from the future head and to the future caudal. Let us take as an example the frog egg. Although in form it is an almost true sphere, we can easily distinguish its upper and lower ends. The upper half of the egg contains a black pigment, and the lower end, an almost white pigment. Upon more detailed study we can easily see that the bulk of the yolk is contained at the lower, the so-called vegetative, pole. At the upper, the so-called animalic, pole there is considerably less of yolk and more of cytoplasm. The nucleus of the egg is also found at the animalic pole. Distinctions between the animalic and the vegetative poles are still more sharply pronounced in the eggs of fishes, birds, and reptiles: here, the embryo is found only at the animalic pole, and all the remaining mass of the egg is taken up with reserves of food substances -- the yolk.

The second axis necessary to produce a plane of bilateral symmetry arises only after fertilization. Let us see how this takes place in the frog egg. An egg that has just been laid and fertilized is bound tightly by membranes and at first is oriented randomly. Its animalic pole can be turned to either side, facing upwards or downwards. (It is reasonable that the egg position in which it is turned animalic pole precisely upwards is possible but not very probable.) Soon after fertilization the membrane somewhat slips off the egg, freeing it and allowing it to be turned about. And in reality the

egg then turns with its heavy (the vegetative) pole downward, and the light (animalic) -- upwards. Since now the egg does not turn, it makes no difference how its orientation determined by the earth's gravity is maintained. However, it is still radially symmetrical: where the dorsal and where the abdominal side of the embryo will be is unknown. Only later, after about an hour following fertilization, does a distinct gray zone, the so-called "gray cycle", form at the interface between the black animalic and the white vegetative hemispheres. It is what determines the future dorsal side of the embryo. From this time the egg changes from radially symmetric to bilaterally symmetric. It now has two axes, running through the center of the egg: the first, joining the animalic and the vegetative poles, and the second, perpendicular to it, joining the equatorial point where the center of the gray cycle lies to the opposing equatorial point (future abdominal side).

What determines this second axis, and what does the whole symmetry plane of the egg mean? Why does the gray cycle appear in one and not the other point of the equator? Perhaps the gray cycle in latent form was in the egg from the very onset of development, even before fertilization? No, this is not so. We can perform an experiment on the fertilization of an egg rotated with its animalic pole upwards. If the egg is tilted to one side, and then is released and allowed to return to its former position, the gray cycle forms exactly on the side which was placed downward. If we tilt the egg once more, even to the other side, the gray cycle forms on the side in which the egg was inclined the last time. (All these experiments are possible only during the first period following fertilization. An hour and a half or an hour later new rotations of the egg can no longer alter the place where the gray cycle appears. The plane of bilateral symmetry is already decisively fixed.)

Thus, it has been shown that it is mainly the plane in which under the action of the force of gravity the egg is turned following its separation from the membrane that determines the plane of bilateral symmetry. Consequently, it arises randomly, since the egg is randomly found lying (or swimming) with a given point of the equator below. This regularity in the development of the frog egg was established by the French scientist Ansel' and Vinterbergh. It was confirmed for sturgeon eggs by the Soviet researchers T. A. Detlaf and A. S. Ginsburg.

However, the problem is far from entirely resolved. It has been shown that if an egg from the very onset, even before fertilization, is placed with its animalic pole upwards, and a rotation, naturally, does not occur, the development will proceed normally just the same. What determines the position of the gray cycle in this case? In the opinion of the French scientists, it depends on the site at which the sperm entered the egg in fertilization. But for sturgeon eggs the sperm enters the egg through special apertures in the animalic pole, the so-called micropyle. In this case the sperm cannot determine the future dorsal or abdominal sides, since

the sperm itself moves along the animalic-vegetative axis. Also unclear is the mechanism by which bilateral symmetry is established when the egg is rotated. Perhaps the determining factor is the path of the overflow by a lighter or by a heavier fluid? It is possible that in rotation this path is moved upwards or downwards along the surface of the egg from the inside and those changes which occur on this side determine where the gray cycle appears, and then the orientation in space. And perhaps, as several scientists believe, rotation results in very weak stresses on one side of the egg, and they prove to be sufficient to determine the plane of symmetry.

Above all, it is first of all necessary to explain what role in this process the forces of earth's gravity play, whether they are necessary for the development of the embryo at the early stage. We need experiments in which development would begin in a state of weightlessness. It must not be thought that setting up these experiments is necessary to solve only one particular problem of embryology. Determination of bilateral symmetry is a part of one of the most pressing problems in that why and how from one cell emerge -- are differentiated -- the cells of various organs and tissues differing so widely from each other.

What has been accomplished today? The first task was to find out whether in general development can occur in a space ship and how the unavoidable factors of flight -- overloading and vibration -- affect development. For this purpose in our laboratory preliminary model experiments were conducted, which examined individually the effect of overloading and vibration on the development of various embryos and on the early stages. It was shown that those accelerations and vibrations which accompany liftoff and reentry do not exert marked damaging effects. The following step in the study consisted of experiments in spaceships. One of the first objects investigated by us were eggs of the parasitic worms -- nematodes. They proved to be convenient in several ways, primarily in that they are capable of surviving in a very broad temperature range. This made possible the placing on spaceships of chilled, undeveloping eggs. Under flight conditions at normal temperature they passed through definite stages of development, and following reentry they were chilled again and in this form transferred to the laboratory. There we could observe how their development following the flight transpired. As was reported in a previous account on the results of the flights of the space ships "Vostok-3" and "Vostok-4", the development of embryos under space flight conditions takes place without any marked deviations. But it must be kept in view that in these experiments the effect of the flight on already fertilized eggs was studied and we did not engage in investigating the effect of weightlessness on how the plane of symmetry was situated. This considerably more complex task will be solved during subsequent flights. Also in the schedule are several other embryological investigations related both to solving specific tasks

of space biology necessary to assure the safety of flights and related to important problems of theoretical biology.

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